

WE CLAIM:

1. A system for separating molecules having different charges and capturing a molecule of interest for detection, comprising:

5 a) a microstructure plate comprising:
at least one microstructure, each microstructure comprising a series of microstructure sections and channels, wherein each microstructure section is directly interconnected to at least one other microstructure section by at least one channel, the series comprising:

10 at least one sample accepting microstructure section, wherein the sample accepting section is fluidly connected to the exterior of the microstructure plate;

at least one first electrode microstructure section;

at least one second electrode microstructure section;

15 at least one capture microstructure section containing a capture matrix, wherein the capture microstructure section is between the first and second electrode microstructure sections in the series;

wherein the microstructures in the microstructure plate are formed by at least two layers of material, wherein at least one layer is a sealing plate layer which seals at least one channel or microstructure section in the assembled microstructure plate; and

20 b) an electrode assembly, the electrode assembly having at least one first and at least one second electrode, wherein each first electrode microstructure section is in electrical contact with at least one first electrode, and wherein each second electrode microstructure section is in electrical contact with at least one second electrode.

- 25 2. The system of claim 1 wherein the sealing plate comprises at least one opening to the exterior of the microstructure plate.

30 3. The system of claim 2 wherein at least one opening of the sealing plate aligns with at least one sample accepting microstructure section.

4. The system of claim 2 wherein at least one opening of the sealing plate aligns with at least one electrode microstructure section.
5. The system of claim 4 wherein each electrode of the electrode assembly extends through at least one opening in the sealing plate towards at least one electrode microstructure section.
6. The system of claim 2 wherein at least one opening of the sealing plate aligns with the capture microstructure section.
7. The system of claim 1 wherein at least one layer of the microstructure plate other than the sealing plate comprises at least one opening to the exterior of the microstructure plate.
8. The system of claim 7 wherein at least one opening of the non-sealing plate layer aligns with at least one sample accepting microstructure section.
9. The system of claim 7 wherein at least one opening of the non-sealing plate layer aligns with at least one electrode microstructure section.
10. The system of claim 9 wherein each electrode of the electrode assembly extends through at least one opening in the non-sealing plate layer towards at least one electrode microstructure section.
11. The system of claim 1 wherein the microstructure unit comprises two capture microstructure sections, wherein one capture microstructure section is positioned in the series between the sample accepting microstructure section and the first electrode microstructure section, and the second capture microstructure section is positioned in the series between the sample accepting microstructure section and the second electrode microstructure section.
12. The system of claim 1 wherein at least one layer of the microstructure plate is transparent to light.
13. The system of claim 1 wherein the capture matrix comprises a material having the ability to covalently or non-covalently bind at least one molecule of interest.
14. The system of claim 13 wherein the capture matrix is positioned within the capture microstructure section so that the molecule of interest travels across the microstructure tangential to the surface of the capture matrix when the first and second electrodes are energized to produce an electric field.

15. The system of claim 13 wherein the capture matrix is positioned within the capture microstructure section so that the molecule of interest travels through the microstructure orthogonal to the surface of the capture matrix when the first and second electrodes are energized to produce an electric field.
- 5 16. The system of claim 13 wherein the capture matrix binds the molecule of interest specifically.
17. The system of claim 16 wherein the capture matrix comprises an affinity binding material selected from the group consisting of antibodies, streptavidin and avidin.
- 10 18. The system of claim 17 wherein the capture matrix binds the molecule of interest non-specifically.
19. The system of claim 18 wherein the capture matrix comprises a material selected from the group consisting of metal chelate resins, anionic resins, cationic resins, polyvinylidene fluoride, nitrocellulose, and positively charged nylon.
- 15 20. The system of claim 1 wherein the capture matrix impedes the movement of a molecule of interest.
21. The system of claim 20 wherein the capture matrix comprises a material selected from the group consisting of cellulose, glass fiber, nylon, and hydrogels.
22. The system of claim 21 wherein the capture matrix is a hydrogel selected from the group consisting of agarose, polyacrylamide, aminopropylmethacrylamide, 3-sulfopropyl-3-(dimethylammonio)propylmethacrylate, methacrylic acid, 3-sulfopropylmethacrylate potassium salt, glycerylmonomethacrylate, and derivatives thereof.
- 20 23. The system of claim 20 wherein the capture matrix is positioned within the capture microstructure section so that the molecule of interest travels through the microstructure orthogonal to the surface of the capture matrix when the first and second electrodes are energized to produce an electric field.
- 25 24. The system of claim 1 wherein at least two channels connecting at least three microstructure sections lie in a three-dimensional configuration.
25. The system of claim 1 wherein the channels connecting the microstructure sections lie in a substantially planar configuration.
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26. The system of claim 1 wherein the microstructure plate is comprised of more than two layers of material, the layers comprising a plurality of voids which define the microstructure sections and channels when the layers are aligned.

27. The system of claim 26 wherein the voids defining the channels of the microstructure lie within a single layer.

28. The system of claim 26 wherein the voids defining the channels of the microstructure lie within more than one layer.

29. The system of claim 26 wherein the capture matrix is held between two layers in order to position it within the capture microstructure section.

30. The system of claim 20 wherein at least one layer is formed from a self-sealing material.

31. The system of claim 30 wherein the self-sealing material is polydimethylsiloxane.

32. The system of claim 20 wherein at least one layer is formed from polytetrafluoroethylene.

33. The system of claim 1 wherein the channels between the microstructure sections of the microstructure have a cross-sectional area between 10,000 and 9,000,000 μm^2 .

34. The system of claim 1 wherein the channels between the microstructure sections of the microstructure have a cross-sectional area between 10,000 and 250,000 μm^2 .

35. The system of claim 1 wherein the channels between the microstructure sections of the microstructure have a cross-sectional area between 25,000 and 250,000 μm^2 .

36. The system of claim 1 wherein the microstructure plate is approximately 8.5 cm by 11 cm.

37. The system of claim 36 wherein the microstructure plate comprises a plurality of rectangularly arrayed microstructures.

38. The system of claim 37 wherein the microstructure plate comprises 96 rectangularly arrayed microstructures.

39. The system of claim 37 wherein the microstructure plate comprises 384 rectangularly arrayed microstructures.

40. The system of claim 37 wherein the microstructure plate comprises 1536 rectangularly arrayed microstructures.

41. The system of claim 37 wherein the electrode assembly comprises 192 regularly arrayed sets of first and second electrodes.
42. The system of claim 37 wherein the electrode assembly comprises 768 regularly arrayed sets of first and second electrodes.
- 5 43. The system of claim 37 wherein the electrode assembly comprises 3072 regularly arrayed sets of first and second electrodes.
44. The system of claim 1 wherein the electrode assembly is integrated within the material of the microstructure plate.
45. The system of claim 44 wherein the electrode assembly is embedded within the sealing
10 plate.
46. The system of claim 44 wherein the electrode assembly is a printed circuit on the sealing plate.
47. The system of claim 44 wherein the electrode assembly is held between two layers of the microstructure plate.
- 15 48. The system of claim 1 wherein the electrode assembly comprises an electrode support plate formed from a rigid or semi-rigid material, the electrodes being fixedly held on or within the electrode support plate.
49. The system of claim 1 wherein each pair of first and second electrodes in the electrode assembly is controlled individually.
- 20 50. The system of claim 1 wherein all first electrodes in the electrode assembly and all second electrodes in the electrode assembly are controlled together.
51. A method for separating molecules having different charges utilizing the device of claim 1, comprising the steps of :
- 25 (a) filling the microstructure with a liquid,
- (b) introducing a sample into a sample-accepting microstructure section of the apparatus,
- (c) energizing the electrode assembly for a sufficient period of time to allow a charged molecule of interest in the sample to migrate towards an electrode of the electrode assembly and to be caught in
30 the capture matrix, and

(d) detecting the charged molecule of interest caught in the capture matrix.

52. The method of claim 51, further comprising the preparatory step of placing at least one capture matrix in at least one capture microstructure section of a microstructure of the apparatus of claim 1.

53. The method of claim 52 wherein the capture matrix is a hydrogel matrix.

54. The method of claim 53 wherein the hydrogel is polymerized after being placed in the capture microstructure section, the method further comprising the step of subjecting a hydrogel precursor to UV irradiation after being placed in the capture microstructure section.

55. The method of claim 51 wherein the capture matrix is a membrane.

56. The method of claim 51 wherein the liquid is an aqueous buffer.

57. The method of claim 56 wherein the aqueous buffer is selected from the group consisting of: Tris hydrochloride buffers, Tris borate buffers, histidine buffer, β -alanine buffers, adipic dihydrazide buffers, and HEPES buffers.

58. The method of claim 56 wherein the microstructure is filled with an aqueous buffer by introducing the buffer into the sample microstructure section under pressure.

59. The method of claim 51 wherein the microstructure is filled with liquid by an automated pipettor.

60. The method of claim 51 wherein the sample is introduced into the sample-accepting microstructure section by an automated sample transfer device, and wherein the sample is transferred from the well of a microtiter plate.

61. The method of claim 51 wherein the electrode assembly is separate from the microstructure plate in step (c), further comprising the step of lowering the electrode assembly into electrical contact with the first and second microstructure sections before energizing the electrode assembly in step (d).

62. The method of claim 51 wherein the electrode assembly is in place when the sample is loaded into the sample-accepting microstructure section in step (c).

63. The method of claim 51 wherein the electrodes are energized to apply 1 μ Amp to 10 mAmp of current through the microstructure.

64. The method of claim 51 wherein the electrodes are energized to apply 1 μ Amp to 5 mAmp of current through the microstructure.
65. The method of claim 51 wherein the electrodes are energized to apply 5 μ Amp to 1 mAmp of current through the microstructure.
- 5 66. The method of claim 51 wherein the electrodes are energized to apply a potential of 0.1 V to 500 V across the microstructure.
67. The method of claim 51 wherein the electrodes are energized to apply a potential of 0.5 V to 100 V across the microstructure.
68. The method of claim 51 wherein the electrodes are energized to apply a potential of 10 1.0 V to 40 V across the microstructure.
69. The method of claim 51 wherein the detection in step (e) is by a method selected from the group consisting fluorometry, colorimetry, luminometry, mass spectrometry, electrochemical detection, and radioactivity detection.
70. The method of claim 69 wherein the detection in step (e) is by fluorometry.
- 15 71. The method of claim 69 wherein the charged molecule of interest is detected by placing at least a portion of the apparatus containing the microstructure plate into a microtiter plate reader.
72. The method of claim 51 wherein the charged molecule of interest is the product of a substrate reaction wherein the net charge of a substrate is changed in the enzymatic 20 reaction.
73. The method of claim 72 wherein the charged molecule of interest and the substrate both comprise a detectable labeling moiety.
74. The method of claim 73 wherein the labeling moiety is a fluorescent moiety.
75. The method of claim 72 wherein the method is capable of detecting the enzymatic 25 conversion of at least 10% of the substrate.
76. The method of claim 72 wherein the method is capable of detecting the enzymatic conversion of at least 1.0% of the substrate.
77. The method of claim 72 wherein the method is capable of detecting the enzymatic conversion of at least 0.1% of the substrate.

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